LETHAL EFFECTS OF ACIDIFIED SEAWATER ON *PENAEUS MONODON* AND THE INTERACTION OF SALINITY AND PH ON SUB-LETHAL EFFECTS

Acidification commonly occurs in marine aquaculture systems when ponds are built using acid-sulphate soils containing pyrite (Boyd, 1982). Low salinity can accompany low pH in situations where run-off from acid-sulphate sediments in pond dykes enters ponds during heavy rains.

Low pH can predispose prawns to disease and influence the toxicity of other toxins, eg. ammonia and aluminium, and in acid waters crustaceans may experience impaired ionic regulation (Morgan and McMahon, 1982). The aims of this study were to estimate lethal and 'minimum acceptable' levels of low pH for *Penaeus monodon* in acidified water and to investigate the interactive effects of low pH and salinity on prawn weight gain, moulting frequency, dry matter content and haemolymph osmotic pressure.

Static bioassays were conducted in 70 L aquaria with three replicate aquaria, each containing 10 prawns, for all low pH treatments. pH was adjusted using 10 N HCl and all aquaria were lightly aerated (100 mL min^{-1}) to maintain dissolved oxygen levels above 5.0 mg O_2 L^{-1}. The average individual initial prawn weight was between 4 and 6 g for all experiments. To estimate lethal levels a bioassay was run for 96 h with pH levels of 7.8, 7.0, 6.1, 5.1, 4.1, 3.8 and 3.0. Prawns survived well (>90%) at pH levels of 5.1 or above. The 96 h LC_{50} (95% confidence limits) estimated was 3.7 (3.4, 4.1).

To assess sub-lethal effects a longer term (23 d) growth experiment was conducted. Eight treatments were established; six at 30 ppt with average pH values of 7.8, 7.3, 6.7, 6.1, 5.5 and 4.9 and two others at 15 ppt with average pH values of 7.8 and 5.5. The effects of treatment on survival rate were not significant ($P>0.05$). Other performance data were analysed using single factor ANOVA (including all eight treatments) or using two factor ANOVA (including data from treatments at pH levels 7.8 and 5.5 for both salinities, 15 and 30 ppt. Growth was depressed at pH ≤ 5.5 at 30 ppt ($P<0.05$) and although salinity did not affect growth ($P>0.05$) there was a significant pH/salinity interaction ($P<0.05$). The absence of a significant difference in growth between 15 and 30 ppt was surprising as prawn farmers in Australia and overseas place great emphasis on maintaining low salinity levels in *P. monodon* ponds. This does not appear necessary in terms of the physiological requirements of *P. monodon* although low salinity could influence other biotic components of pond ecosystems.

The 'minimum acceptable' level was defined as that level which reduced growth by 5% (the EC_5) and was estimated, using two-phase linear regression analysis (Sedgwick, 1979), as being 5.9 pH units for *P. monodon* at a salinity of 30 ppt.

Moulting frequency was highest at pH 4.9 (30 ppt) and was inversely related to salinity, while the interaction was not significant ($P>0.05$). The dry matter content was depressed at pH 4.9 (30 ppt) but unaffected by salinity or the interaction ($P>0.05$)

Juvenile *Penaeus monodon* are efficient osmoregulators in the range 15 to 30 ppt with an isosmotic point of between 23 and 25 ppt (Cawthorne *et al.*, 1983). However, a reduction in internal osmolarity at reduced pH had been recorded for a number of freshwater crustaceans and fish (Morgan and McMahon, 1982; Hobe *et al.*, 1983). To investigate whether changes in osmotic pressure might explain the pH/salinity interaction prawns were exposed to combinations of two pH (7.8 and 5.6) and salinity (15 and 30 ppt) levels for three days; sufficient time for osmotic and ionic equilibrium to be reached. At the end of the experiment osmotic pressures in the water and prawn haemolymph were measured and the difference between these two values (Dop) calculated as an indication of osmoregulatory ability. At both salinities (15 and 30 ppt) haemolymph osmotic pressure was closer to ambient osmotic pressure at reduced pH (5.6). Both salinity ($P<0.001$) and pH ($P<0.01$) significantly reduced Dop, and there was no interaction ($P>0.05$). Although the results of this experiment showed that reduced pH lowered osmoregulatory ability in *Penaeus monodon*, they did not confirm the hypothesis that differences in osmoregulatory ability were responsible for the interaction between pH and salinity on weight gain. The interactive effects of pH and salinity on ionic regulation may warrant further investigation.

The estimation of lethal and sublethal low pH levels for *P. monodon* (3.8 and 5.9 pH units respectively) should assist prawn farmers with the management of acidic ponds.

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